

## Effect of adding Nano-NPK fertilizer and humic acid on some vegetative growth characteristics and active components of Myrtle *Myrtus communis* L.

Laila Turki Fadalalah\*, Ghufuran Zaid Khalaf Al-Rikabi and Hadiyah A. Atiyah

Department of horticulture - College of Agriculture - University of Thiqr, Iraq

\*Corresponding author's e-mail: [lailaturky8@gail.com](mailto:lailaturky8@gail.com)

The current study was conducted during the 2019–2020 growing season in Dhi-Qar University's Alsaran Canopy. Nano fertilizer was used to test the effects of adding Nano-<sup>1</sup> macro elements NPK at three concentrations (0, 30, 60) mg l<sup>-1</sup> on some growth characteristics of Myrtle plant, *Myrtus communis*. Three replicates of the experiment were used in the RCBD design. Important Difference Test at 0.05 probability. Results appear that The Nano fertilizer is 60mg l<sup>-1</sup> significantly increased the average plant height, stem diameter, leaf area, chlorophyll and carbohydrates leaf content, and percentage of oil, fresh weight. The concentration of humic acid Significantly increasing plant height, branch count, stem diameter, chlorophyll content, carbohydrates, and the highest percentage of oil. It recorded an average of (87.44 cm, 12.13 mm, 10.56 branches). 2, 105.44 mg.100 g fresh weight, 82.00 g. dry matter and 0.51%, respectively, while H1 treatment excelled with the largest leaf surface area of 8.79 cm<sup>2</sup>. The interaction treatment H2N1 was superior to the control treatment H0N0 and the rest of the treatments in the study regarding plant height, stem diameter and leaf area, while H2N2 was superior in fruit content of chlorophyll and carbohydrates.

**Keywords:** Myrtle plant, nano fertilizer, humic acid, chlorophyll, carbohydrates.

### INTRODUCTION

The Myrtle plant, *Myrtus communis* L., belongs to the family Myrtaceae. It belongs to the perennial plant family. It is one of most important plants for green plant hedges, including 150 genuses and more than 5,500 species naturally cultivated worldwide. It has an effective role in preserving the purification of the environment and human health. It was used as food in ancient times and in improving the value and flavor of food because it contains many biologically effective compounds that are important in manufacturing cosmetics (Jamshidi-Kia *et al.*, 2018).

Nano fertilizer of NPK is an essential nutrient to nourish plants and increase their production. One of the vital minerals that plants require in significant amounts is nitrogen. It is the engine of plant growth and has many physiological roles in plants, including its entry into the formation of protoplasm and compounds, proteins, nucleic acids, and amino acids and the production of chlorophyll are all essential to the health of the plant (Kafeel *et al.*, 2023). As well as, plants require significant amounts of phosphorus, as one of the necessary elements. Which also plays crucial physiological roles in plants (Malhotra *et al.*, 2018).

It enters as one of the components of the cell, enters into the synthesis of organic substances in plant tissues such as phytin, phospholipids and phospholipids, they have a direct connection to the respiration process and are involved in the synthesis of nucleic acids, which has a direct connection to the processes of cell division, meristematic growth, and the synthesis of energy-containing substances (Malhotra *et al.*, 2018).

Potassium is one of the essential nutrients that plants need in large quantities, and it is the only element found in an ionic form in plant cell juice K<sup>+</sup>. In order to increase the effectiveness of photosynthesis, potassium is crucial, thus, increasing amount of carbohydrates formed. Potassium is concentrated in the growing and meristematic tops of the plant, which suggests that it has a relationship with the process of cell division (Mengel *et al.*, 2001).

The main purpose of using humic acid is to improve the plant's nutritional status and, thus, increase production as a natural fertilizer alternative to industrial fertilizers (Al-Tamimi, 2021). Mustafa *et al.* (2018) found that when spraying fig seedlings NPK nano fertilizer with 100,200,300,400 mg.L<sup>-1</sup> concentrations, the concentration of 400 mg.L<sup>-1</sup> gave a highest percentage of chlorophyll in

reached 46.6 mg.L<sup>-1</sup>, while control treatment contained a concentration of (500 mg.L<sup>-1</sup>) of the regular NPK fertilizer, amounted to 45.9 mg. L<sup>-1</sup>. Daoud (2020) explained that when using NPK nano fertilizer on pomegranate seedlings, it showed significant superiority in most of the study indicators, which led to an increase in stem diameter, stem length, branches number, leaves number, leaf area, carbohydrates, chlorophyll, and fresh weight. EL- Ghamry *et al.* (2009) showed that adding humic acid, enhanced chlorophyll a, chlorophyll b, and carotenoids, either alone or in conjunction with amino acids, in bean leaves. applying humic acid to cucumber plants in Babylon at a dosage of 6 mL.L<sup>-1</sup> significantly improved their vegetative development properties (plant height, branches number, leaves number; Yousin, 2011).

Based on critical information regarding special requirements of plants for special fertilizers, The study aimed to know effect of different concentrations of NPK nano fertilizer and humic acid in improving vegetative growth characteristics and increasing the chemically active substances and the Myrtle plant's oil percentage.

## MATERIALS AND METHODS

An experiment was carried out in the wooden canopy of the College of Agriculture and Marshes-University of Dhi Qar in the agricultural season 2019-2020 in a silty-soil to examine the effects of humic acid and the nano-fertilizer NPK on the growth of the Myrtle plant, *Myrtus communis* L., and its volatile oil content. The plants were two years old at that time. A local selection from one of the private nurseries in Nasiriyah. On January 9, 2019, the plants were placed in plastic containers that were 17 cm in height by 23 cm in diameter and were filled with a sterile growing media made of sand and peat moss. Each seedling has 5 main branches left after all the plants were cut to a height of 55 cm. The factorial experiment was designed using the RCBD design and with three replications. To evaluate some of the chemical and physical features of the soil, samples from the study soil were taken and then mixed, as shown in the Table 1.

**Studied parameters:** Plant height (cm): measured from growth medium's surface to plant's apex using only a metric tape. Stem diameter (mm): was measured at the height of 5 cm from the surface of the growing medium using a Vernier's foot. Leaf area (cm<sup>2</sup>): was measured by the METER AREA LASER 202-C device created by C I D. BIO-Science, an American business. The carbohydrate content of the leaves: determined by the method of phenol-sulfuric acid-phenol according to Dubois *et al.*(1956). Chlorophyll leaves content (mg.100 g fresh weight): 1 g of leaves was taken and crushed with 10 ml of acetone, then placed in a centrifuge for 5 minutes at a speed of 3000 revolutions/min. Spectrum readings for wavelengths 663 and 665 nm were recorded

using a spectrophotometer. The total chlorophyll is estimated by using the following equation:

$$\text{Total chlorophyll (mg/L)} = 20.2 \times (665) D \times 8.02 + (645) D$$

The percentage of volatile oil%: was estimated according to the method mentioned by Keverson *et al.* (2009)

$$\text{Percentage of volatile oil \%} = \frac{\text{weight of the resulting oil (mg)}}{\text{sample weight of the leaves (g)} \times 100}$$

**Table 1. Physical, and chemical properties of soil.**

Type of analysis	Unit of measure	soil	Method
EC	dS.m <sup>-1</sup>	4.7	Page <i>et al.</i> , (1982)
pH	----	7.20	
Total nitrogen	mg. kg <sup>-1</sup>	15.4	
R phosphorous	mg. kg <sup>-1</sup>	11.45	
R Potassium	meq. l <sup>-1</sup>	22.2	
Organic matter	g.kg <sup>-1</sup>	10.00	Black (1965)
Percentage of clay	%	40.2	
Percentage of silt	%	10.3	
Percentage of sand	%	49.5	
Soil texture	---	sandy clay	

## RESULTS AND DISCUSSION

**Plant height (cm):** According to the statistical analysis, there were significant differences between the averages of the treatments that included adding humic acid and NPK nano fertilizer in terms of the characteristic of plant height. The plants treated with humic acid generated the highest height of 87.44 cm in compared to the control plants, which had the lowest height of 75.22 cm.

**Table 2. Effect of adding NPK Nano fertilizer and humic acid on plant height (cm)**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect of humic acid	The effect of the NPK nano fertilizer
	2	1	0		
0	92.33	79.00	54.33	75.22	69.11
30	101.33	88.33	72.00	87.22	83.89
60	97.00	84.33	81.00	87.44	96.89
L.S.D0.05		6.618		3.821	3.821

**Stem diameter:** The results in table 3 clearly demonstrated that the plants given NPK nano fertilizer at a dose of 60 mg



were much superior. By producing stems with a diameter of 12.36 mm, L 1 outperformed control plants, which produced stems with a diameter of 9.55 mm. the 2 mg of humic acid that was applied to the plants. L1 significantly outperformed plants treated with a dose of 1 mg.L1 and the control plants, producing stems with a diameter of 12.13 mm as opposed to the control plants' stems with a diameter of 9.39 mm. The results also revealed substantial variations in the two- way interaction averages, with N1H2 plants producing stems with an average diameter of 13.03 mm and 6.17 mm respectively, and N0H0 plants.

**Table 3. Effect of adding NPK Nano fertilizer, humic acid, and their interactions on stem diameter (mm)**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect of humic acid	The effect of the NPK nano fertilizer
	2	1	0		
0	12.32	9.68	6.17	9.55	9.39
30	13.03	10.29	10.93	11.02	11.42
60	11.73	13.10	11.54	12.36	12.13
L.S.D0.05		1.424		0.822	0.822

**Leaf area:** Table 5 showed that increasing leaf area with the addition of NPK and humic acid nano fertilizer was significantly superior, as evidenced by the maximum leaf area at 8.48 cm<sup>2</sup> from a plants given NPK nano fertilizer at a dosage of 60 mg. Compared to control plants, L-1 reported a lowest leaf area, measuring 6.96 cm<sup>2</sup>, The Table's also showed that humic acid at a concentration of 1 mg. Compared to the control plants, L 1 had the largest average leaf area, 8.79 cm<sup>2</sup>, and lowest leaf area, 6.80 cm<sup>2</sup>. Treated N1H2 plants performed exceptionally well, with the maximum average leaf area reaching 9.57 cm<sup>2</sup>, compared to the lowest area produced by N0H0 plants, which was 4.77 cm<sup>2</sup>. This indicates that the binary interaction had a considerable impact.

**Table 4. Effect of adding NPK Nano fertilizer, and humic acid and their interactions on leaf area (cm<sup>2</sup>.plant)**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect of humic acid	The effect of the NPK nano fertilizer
	2	1	0		
0	6.93	8.70	4.77	6.96	6.80
30	9.57	8.97	7.83	8.37	8.79
60	8.93	7.43	8.29	8.48	8.22
L.S.D0.05		0.535		0.309	0.309

**Chlorophyll leaves content mg 100 g<sup>-1</sup> of Fresh weight:**

Table (6) of the data showed that there were substantial changes at some times. The results showed a significant differences between the treatments, with the addition of NPK nano fertilizer being significantly better than the addition of

N2, and the highest content of chlorophyll being 96.89 mg 100 g<sup>-1</sup> of fresh weight, compared to control treatment which gave lowest chlorophyll content of 78.56, and the plants of the H2 treatment also excelling by giving a highest chlorophyll content.

**Table 5. Effect of adding NPK Nano fertilizer, humic acid, and their interactions on chlorophyll content leaves (100 mg<sup>-1</sup> fresh weight)**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect of humic acid	The effect of the NPK nano fertilizer
	2	1	0		
0	83.33	73.33	42.00	78.56	66.22
30	97.00	94.00	90.33	90.00	93.78
60	110.33	102.67	103.33	96.89	105.44
L.S.D0.05		4.397		2.539	2.539

**Total soluble carbohydrates leaves content of (mg g<sup>-1</sup> dry matter):**

Table 6 demonstrated that the amount of total soluble carbohydrates in the leaves was significantly affected by treatment with humic acid and nano NPK fertilizer. The largest content, 84.56 mg.g<sup>-1</sup> dry matter, was produced by the treated of 60 mg.l. 1 of the NPK nano fertilizer, while the lowest value, 70.67 mg.g<sup>-1</sup> dry matter, was produced by the control plants. Moreover, 2 g of humic acid H2 was applied to the plants. The control plants produced the lowest quantity of carbohydrates, amounting to 74.94 mg g<sup>-1</sup> dry matter, whereas L 1 excelled with the highest content, which came to 82.00 mg g<sup>-1</sup> dry matter. The N2H2 plants had the maximum content of 91.67 mg g<sup>-1</sup> dry matter, while N0H0 plants had the lowest content, 52.67 mg g<sup>-1</sup> dry matter, demonstrating the binary interaction's clear superiority.

**Table 6. Effect of NPK Nano fertilizer, humic acid, and their interactions on total soluble carbohydrates leaves content (mg g<sup>-1</sup> dry matter)**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect of humic acid	The effect of the NPK nano fertilizer
	2	1	0		
0	87.67	84.50	52.67	70.67	74.94
30	74.33	83.67	78.33	80.50	78.78
60	91.67	73.33	81.00	84.56	82.00
L.S.D0.05		5.078		2.932	2.932

**Percentage of volatile oil in leaves %:** Table 8 shows the plants treated with NPK nano fertilizer N2 gave a highest value at 0.49% while the lowest value of 0.36. % produced from N0 plant extracts. Also, H2 treatment plants excelled with a highest oil percentage, which amounted to 0.51%, the lowest content of 0.39% produced from H0 plants, which reached 0.24% recorded from N0H0 plants.



**Table 7. Effect of treatment with NPK Nano fertilizer, humic acid, and their interactions on the percentage of the volatile oil of Myrtle plant (%).**

NPK Nano fertilizer concentrations mg. liter	Humic acid g.L <sup>-1</sup>			The effect Means of the NPK nano fertilizer	The effect of humic acid
	2	1	0		
0	0.48	0.46	0.24	0.36	0.39
30	0.47	0.49	0.43	0.48	0.46
60	0.52	0.51	0.51	0.49	0.51
L.S.D0.05		0.036		0.021	0.021

Tables 2-4 show that there was an increase in studied vegetative characteristics (plant height, number of branches, leaf area, and stem diameter) with an increase in NPK Nano fertilizer concentration. This, in turn, is explained by the fact that the NPK Nano fertilizer increases the plant's efficiency in absorbing water and nutrients. The major elements, play a significant role in plant's various metabolic processes. This results in an increase in growth, which is then positively reflected in the activity of the vegetative system. Growth hormones' action causes cells to expand, increased the branches number and their length, and subsequently an increase in the number of leaves. The outcomes were in agreement with (Buzea *et al.* 2007; Alhasany *et al.*, 2019). The fact that nano-fertilizers increase the availability of available nutrients to the plant over a longer time period and with a release consistent with plant growth is the cause of the considerable concentration of chlorophyll and carbohydrates in tables (6-7), according to Buzea *et al.* (2007) and Seleiman *et al.* (2020). As a result, the rate of photosynthesis and the production of chlorophyll both rise, hence accelerating total growth (Raab and Terry, 2007). Which in turn increases the concentration of total carbohydrates. The function of main elements, particularly nitrogen, which regulates the activity of growth hormones, cytokines, and auxins, which play a large role in the course of the numerous metabolic processes of the plant, is responsible for the significant rise in carbohydrates. These findings concur with those of earlier research (Matrood & Al-Taie, 2017). The oil percentage has increased significantly, as seen in Table (8). The rise may be caused by phosphorus's increased availability of its direct incorporation into creation of sinter compounds or as a catalyst during their production (Tinter, 2008).

Phosphorus additionally increases, manufactures, and accumulates carbohydrates, which boosts secondary chemical formation, including essential oils. Nitrogen fertilizer from oil increases vegetative growth and oil extraction (Al-Samarrai, 2000). This investigation confirms Boyle and others (1991) that nitrogen fertilizer increased volatile oil in the desperate plant. The height branches number, stem diameter, and a leaf area of plants treated with humic acid may increase because organic fertilizers increase nitrogen levels in the plant, which increases chlorophyll in the leaves, encourages growth, increases cell size, and increases the vegetative system and

leaf area (Boaissa and Ghayath, 2006; Altai *et al.*, 2020). When treated with humic acid, leaves have more chlorophyll due to its role in processing large amounts of nitrogen that enters the formation of chlorophyll. It also increases carbon fixation as an organic fertilizer, which increases the plant's ability to supply the car. Organic matter prepares nitrogen and magnesium, which are significant since they are in the heart of the chlorophyll molecule. These results agreed with what was found (Al-Tamimi, 2012). The improvement in vegetative growth indicators given in tables (2,4 and 3,5) may be related to the rise in soil absorption of nutrients and water, the activity of photosynthesis and the manufacturing and transfer of nutrients to all areas of the plant, and the increase in leaf element percentage (Al Sahhaf, 1989). As each researcher confirmed (Peter and Carl, 2005), nitrogen builds the chlorophyll molecule, and phosphorus boosts roots and absorption efficiency. Potassium builds carbohydrates, iron, manganese, and magnesium build the chlorophyll molecule, and boron preserves the pigment by increasing the activity and effectiveness of some growth hormones, especially cytokinin and alkanetin, which increase plant green pigments and photosynthesis in leaves, which increases nutrient production and transfer to plant parts. Due to increased leaf area (Table 5) and chlorophyll concentration (Table 6), photosynthesis and its outputs (carbohydrates) may rise humic acid equips roots (Table 7). Present findings are in agreement with Khaled and fawy, (2011) who stated that carbs, amino acids, and proteins boost growth of plant.

**Conclusion:** The study found that 60 mg l-h of NPK nano fertilizer improved plant growth and the content of vegetative, chemical, and oil. In most vegetative growth, chemical component and oil percentage characteristics. Humic acid-treated plants have a concentration of 2 g l-1.

**Author's Contributions statement:** Laila T. Fadalah, Contributed to the idea, field implementation, and writing. Ghufraan Z. Khalaf AL\_Rikabi contribute to data collection and part of the analysis. Hadiyah A. Atiyah contributes to fieldwork and linguistic and statistical review.

**Conflict of interest:** Laila T. Fadalah and other authors declare no conflicts of interest

**Acknowledgement:** Laila T. Fadalah, I cannot express enough thanks to my team in this study for their continued support and encouragement.

**Funding:** Not applicable.

**Ethical statement:** This article contains no studies with human participants or animals performed by authors.

**Availability of data and material:** That manuscript has never been published and is not under consideration for publication elsewhere.



**Code Availability:** Not applicable.

**Consent to participate:** All authors are participating in this research study.

**Consent for publication:** All authors are giving their consent to publish this research article in JGIAS.

## REFERENCES

- Al-Hasan, I. I. S. 2021. The effect of spraying with some stimuli on the growth of the Myrtle plant (*Myrtus communis* L.), its volatile oil content and its active compounds. PhD Thesis - College of Agriculture - University of Basra.
- Alhasany, A.R., D.S.K. Altai, and A.H. Noaema. 2019. Effect of Foliar Nano-Fertilizers of Marine Algae Extract and Boron on Growth and Yield of Faba Bean (*Vicia faba* L.). Indian Journal of Ecology 46:251-253
- Al-Sahhaf, F. H. 1989. Applied plant nutrition, Dar Al-Hikma Press, Ministry of Higher Education and Scientific Research, Iraq; pp 258
- Al-Samarrai, I. S. M. 2000. The effect of nitrogen fertilization on the growth of lemongrass, the quantity and quality of volatile oil, and the effect of the oil on the growth of some fungi. PhD Thesis, College of Science, University of Baghdad, Iraq.
- Altai, D.S.K., A.R. Alhasany, and K.A.K. Al Tameemi. 2020. Role of Humic Acid and Amino Acids in Increasing Growth and Productivity of Mungbean Varieties Grown under Newly Reclaimed Soil. Indian Journal of Ecology 47 : 11-16.
- Al-Tamimi, Nasser Habib Mahbis. 2021. The effect of mycorrhiza fungus, humic fulvic acid and licorice extract on the morphological and physiological characteristics of the okra plant.
- Black, C.A. 1965. Method of Soil Analysis. Part(1). Physical properties. Am. Soc. Agron. Inc. Publisher, Madison, Wisconsin, USA.
- Boyle, T.H., L.E. Craker and J.E. Simon. 1991. The growing medium and fertilization regime influence rosemary growth and essential oil content. HortScience 26: 33-34.
- Buzea C, I. I. Pacheco and K. Robbie. 2007. Nanomaterials and nanoparticles sources and toxicity Biointerphases 2:17-71.
- Daoud, F. K. D. 2020. Response of pomegranate seedlings to balanced NPK nano fertilization and salicylic acid spraying and their effect on growth and phenolic content of leaves. Master Thesis, Dhi Qar University, Iraq.
- Dubois, M., K. A. J. K. Gilles., P. Hamilton., A. Rebers and F. Smith. 1956. Colorimetric method for determination of sugars and related Substances. Analytical Chemistry 28:350-356.
- El-Ghamry, AM. , K. M. Abd El-Hai and K. M Ghonem. 2009. Amino and humic acids promote growth, yield and disease resistance. Naba bean is cultivated in clay soil. Australian Journal of Basic and Applied Science 3:731\_739.
- Jamshidi-Kia, F., Z. Lorigooini, and H. Amini-Khoei. 2018. Medicinal plants: History and future perspective. Journal of Herbmed Pharmacology 7:1-7.
- Kafeel, U., Jahan, U., and Khan, F. A. 2023. Role of mineral nutrients in biological nitrogen fixation. In Sustainable Plant Nutrition Academic Press, pp. 87-106.
- Keverson, Z.S., C. Natasa., H. Harabovsk and B. SaKac. 2009. Essential Oil Composition of fresh and dried Pepper fruit *Capsicum annum* L. for ground paprika product processing. Quality and safety 1:229-233
- Khaled, H. and H. A. Fawy. 2011. Effect of different levels of humic acids on the nutrient content: plant Growth Soil Properties under Conditions of Salinity. Soli and Water Res 6:21-29.
- Malhotra, H., S Sharma and R. Pandey. 2018. Phosphorus nutrition: plant growth in response to deficiency and excess. Plant nutrients and abiotic stress tolerance. Springer, Singapore. pp.171-190.
- Matrood, A. A and A. H. Al-Taie. 2017. Inhibition Activity of mycorrhizal Fungi *Glomus mosseae* and *G. intradicas* with *Trichoderma harizanum* Against *Rhizoctonia solani* in Okra Plant *Abelmoschus esculentus* (L.). Basrah Journal of Agricultural Sciences 30:72-82.
- Mengel, K., E. A. Kirkby., H. Kosegarten and T. Appel. 2001. Principles Plant Nutrition. Kluwer Academic Publishers
- Mustafa, N.S., H. H. stairway., M.F. El-Dahshouri., M.F and A.S. Mahfouze. 2018. Impact of nano-fertilizer on different aspects of growth performance, nutrient status and some enzyme activities of (Sultani) fig cultivar. Bioscience research 15:29-34.
- Page, A. L., R. H. Miller, and D. R. Keeney. 1982. Method of soil and analysis Part 2, 2nd ed, Agron. 9. Publisher, Madison, Wisconsin, USA.
- Peter, M.B. and C.J. Rosen. 2005. Nutrient cycling and maintaining soil fertility in fruit and vegetable crop systems. Department of Soil, Water and Climate-University of Minnesota. M1193.
- Raab, T. K., and N. Terry. 1994. Nitrogen source regulation of growth and photosynthesis in *Beta vulgaris* L. Plant Physiology 105:1159-1166.
- Seleiman, M. F., K. F. Almutairi, M. Alotaibi, A. Shami, B. A. Alhammad, and M. L. Battaglia. 2020. Nano-fertilization as an emerging fertilization technique: why can modern agriculture benefit from its use? Plants 10: 2.
- Tinter, E. S.J., K. Meisst. and E. Binner. 2008. Influence of input materials and Compositing Operation on Organic Mteer Dinamic Soil Humification. Dinamic plant 1:50 - 59.



Yousin, K. H. 2011. Enhanced on humic acid bio fertilizer (EM\_1) and application methods on growth, flowering yield oN cucumber (*Cucumis natives*). A Thesis

submitted to the college of Agriculture, University of Duhok, Iraq.

